

## **In the Specification**

### **In the Specification:**

Please replace paragraph [0005] with the following amended paragraph:

[0005] HA has been recently applied in the highly molecular of medicine. Generally, it has been reported that HA has functions of (1) ~~nationally~~ **naturally** occurring in body, (2) ~~nonimmunoreaction~~ **no immunoreaction**, (3) degradation and adsorption by human body, (4) mass production and others.

Please replace paragraph [0011] with the following amended paragraph:

[0011] Malson, 1990, U.S. Pat. No. 4,963,666, states a process for producing the carboxyl-containing polysaccharides. A polysaccharide containing carboxyl group **is reacted** with a bi or poly-functional epoxide in an alkaline reagent ~~by dialysis~~. The cross-linking is performed during drying. Malson described the 200 mg of sodium hyaluronate MW  $3 \times 10^6$  were mixed with 6 ml of 0.5% NaOH in a plastic tube and stirred with a glass rod until a clear homogeneous solution has been obtained. Then 2 ml of 1,4-butanediol diglycidyl ether (BDDE) were added and mixed thoroughly. The solution was subjected to shaking overnight and dialyzed against running distilled water for 24 hrs had a weakly acidic pH of about 5.5. The solution was poured into a petri dish of polystyrene and dried at room temperature for 2 days. A transparent, planar, water-insoluble film of 50  $\mu\text{m}$  thickness was obtained.

Please replace paragraph [0012] with the following amended paragraph:

[0012] Sakurai ET. Al., 1987, U.S. Pat. No. 4,716,224, states a cross-linked hyaluronic acid and its use. In the invention, ~~cross-linked hyaluronic acid or salts thereof prepared by cross-linking hyaluronic acid or salts thereof with~~ a poly-functional epoxy compound **is** selected from the group consisting of halomethyloxirane compounds and a bis-epoxy compound selected from the group consisting of 1,2-bis (2,3-epoxypropoxy) ethane, 1,4-bis (2,3-epoxypropoxy) butane, 1,6-

bis (2,3-epoxypropoxy) hexane and a diglycidyl ether of biophenol A or bisphenol F, which has a crosslinking index of 5 to 20 per 100 repeating disaccharides composed of glucuronic acid and N-acetylglucosamine in hyaluronic acid, said cross-linked hyaluronic acid or pharmaceutically acceptable salt thereof being water soluble and stringy.

Please replace paragraph [0013] with the following amended paragraph:

[0013] 10 g of HA solution salt (a molecular weight,  $7.3 \times 10^5$ ) were dissolved in 450 ml of 0.2N sodium hydroxide solution with cooling and the resulting solution was filtered with a 0.45  $\mu\text{m}$  micro-filter. The filtrate was added to 40 ml of 10N sodium hydroxide solution and then 500 ml of ethanol and 6.0 ml of epichlorohydrin with stirring. The reaction was effected at 20° C for 24 hrs and then the reaction mixture was adjusted to pH 6.4 with acetic acid. By addition of 500 ml of ethanol, there was separated a white precipitate, which was then recovered by filtration, washed well with ethanol and dried under reduced pressure. In the other example, 2.0 g of HA sodium salt (a molecular weight,  $2.0 \times 10^6$ ) were dissolved in 100 ml of 0.1N sodium hydroxide under cooling and 100 ml of dioxane and 4.3 g of 1,4-bis (2,3-epoxypropoxy) butane were added. Reaction was effected at 40° C for 2 hrs. The reaction mixture were added to 200 ml of water, the resultant mixture was neutralized with 1N hydrochloric acid and then centrifuged at 300 rpm. The precipitate was washed well with a 1.0M aqueous solution of sodium chloride, and a 0.15M aqueous solution of chloride, dehydrated with ethanol and then dried. Finally, 1.7 g of the cross-linked HA was formed.

Please replace paragraph [0019] with the following amended paragraph:

[0019] De Belder et al., PCT publication No. WO 86/00912, describes a slowly-degradable gel, for preventing tissue adhesions following surgery, prepared by cross-linking a ~~carboxyl-contains~~ carboxyl-containing polysaccharide with a bi-or poly-functional epoxide. The gel ~~with~~ has an infrared absorption frequency of carboxyl group ( $-\text{COOH}$ ) of ester functional group at 1745  $\text{cm}^{-1}$ . 400 mg of sodium hyaluronate molecular weight  $3 \times 10^6$  was dissolved in 4 ml of distilled water for 2 hrs. Then 600 mg of 1,4-butanediol diglycidyl ether (BDDE) was added and admixed thoroughly. HA gel was formed after the 0.15 ml of glacial acetic acid was added and reacted at 60-70° C for 15 hrs.

Please replace paragraph [0020] with the following amended paragraph:

[0020] T. Malson et al., 1986, PCTP publication No. WO 86/00079, states a method for preparing viscous fluid ~~contains~~ **containing** a sterile and pyrogen-free gel of cross-linked hyaluronic acid by reacting hyaluronic acid with the poly-functional epoxide, halohydrin, epihalohydrin or halide.

Please replace paragraph [0027] with the following amended paragraph:

[0027] Balazs et al., 1986, U.S. Pat. No. 4,582,865, states a method for preparing cross-linked gels of hyaluronic acid and products containing such gels. The major characteristic of the invention was that a mixture of sodium hyaluronate and other hydrophilic polymer in a dilute aqueous alkaline solution at a pH of not less than about 9 ~~to a cross-linking reaction~~ **was cross-linked** with divinyl sulfone at about 20° C.

Please replace paragraph [0031] with the following amended paragraph:

[0031] Thompson et al., 1996, U.S. Pat. No. 5,563,186, states a method for preparing cross-linked alginate-based gels for matrix conformance. A composition of matter for matrix conformance formed from an aqueous solution of an alginate polysaccharide and a method of forming an alginate polysaccharide gel **are disclosed**. A Group IIA cation, a dialdehyde, or a diamine may be used to cross-link the resulting alginate gel.

Please replace paragraph [0034] with the following amended paragraph:

[0034] Iguchi et al., 1998, U.S. Pat. No. 5,811,531, states a method for preparing absorbent with stability against salts and process for production thereof. 100 parts of xanthan gum was placed in mixer and stirred with 4 parts of the aqueous of the cross-linking agent obtained by adding of ethylene glycol diglycidyl ether to the methanol solution. The obtained mixture was heated at 140° C for 20 minutes to form the absorbent. The absorbent was obtained using sodium alginate, pectin, and guar gum in the same procedure as above described. G. Hamdi et al., 1998, Journal

of controlled release, Vol 55. page 193-201, states an original method for study in vitro the enzymatic degradation of cross-linked starch micro-spheres. The aqueous phase was prepared by dissolving soluble starch in a 2M sodium hydroxide solution under mechanical stirring. The aqueous phase was pre-emulsified in a cyclohexane-chloroform mixture (4:1,v/v) containing 0.5%(v/v) of sorbitane monooleate. This emulsion was then ~~was~~ added under mechanical agitation at 600 rpm. The reaction was maintained at 40° C for 18 hours. Micro-spheres were then isolated by centrifugation and washed with cyclohexane, extensively with deionized water and finally with ethanol 95%(v/v).

Please replace paragraph [0036] with the following amended paragraph:

[0036] Chitosan was obtained from the deacetylation by using the thermal alkaline. Chitin and chitosan are ~~nationally~~ **naturally** high molecular compounds, with good bio-compatible, biodegradation and almost nontoxin (Dose of lethal, LD<sub>50</sub>=16g/Kg). Besides the different applications on the food industries, the anti-microbial effect of chitosan can be also applied to the encapsulating and pharmaceuticals.

Please replace paragraph [0037] with the following amended paragraph:

[0037] The used chitosan is low impact to the environment, **has** one or more other beneficial properties such as bio-compatible and biodegradation, which ~~are~~ make them suitable for many applications, such as the encapsulating materials, agricultural use, biomedical materials and pharmaceuticals for a wide variety of use.

Please replace paragraph [0038] with the following amended paragraph:

[0038] Unger et al., 1996, U.S. Pat. No. 5,525,710, states a method for preparing highly porous chitosan bodies. Chitosan flakes were dissolved in a solution of dilute acetic acid, and the viscous solution was centrifuged to remove air bubbles. The gel was then made by contacting the viscous hydrocolloid with a solution of sodium hydroxide for 24 hours. The gel was then sliced and immersed into the toluene solution, and cross-linked with 2,4-TDI. The cross-linked product was then dried in a vacuum oven, and ground to ~~a~~ **a** powder, which contained highly

pore volume of the cross-linked chitosan.

Please replace paragraph [0041] with the following amended paragraph:

[0041] Sakurai et al., 1989, U.S. Pat. No. 4, 863,907, states a method for preparing cross-linked glycosaminoglycans and their use. As the example of the invention described, to a mixture of a 12.5% solution of ChS--C sodium salt ~~in~~ and a 0.75N aqueous solution of NaOH was added 1 volume of ethanol under stirring and the resultant sticky precipitate was separated and recovered. This sticky precipitate was added to the epichlorohydrin, the resulting mixture was kneaded well and then it was allowed to stand at 20° C for 24 hours. The ChS--C sodium salt was synthesized in the reaction.

Please replace paragraph [0050] with the following amended paragraph:

[0050] The preferred dry solid ~~content~~ content of hydroxyl-containing polysaccharide in the reaction is 0.2% to 10%. The hydroxyl-containing polysaccharide is chosen from hyaluronic acid, carboxymethyl cellulose, starch, alginate, chondroitin-4-sulfate, chondroitin-6-sulfate, xanthane gum, chitosan, pectin, agar, carrageenan or guar gum. The hydroxyl-containing polysaccharides that can cross-link with the epoxide are all included in this invention. They are not construed to be limiting the scope of this invention.

Please replace paragraph [0067] with the following amended paragraph:

[0067] Sodium hyaluronate (100 mg; 1 molar equivalent of hydroxyl group) was dissolved in 10 ml of distilled water. The 1% dry solid content of HA solution was formed under stirring at room temperature. ~~Following~~ Next, adjustment of the pH of the HA solution was performed by addition of 1.0 N NaOH or 1.0N HCl. These results are showed in the following table (1A).

Please replace paragraph [0071] with the following amended paragraph:

[0071] Sodium hyaluronate (100 mg; 1 molar equivalent of hydroxyl group) was dissolved in 10 ml of distilled water. The 1% dry solid content of HA solution was formed under stirring at

room temperature. ~~Following~~ Next, adjustment of the pH of the HA solution was performed by addition of 1.0 N NaOH or 1.0N HCl. These results were showed in the following table 2. Then EGDGE (224 mg; 2 molar equivalent of hydroxyl group) was added in the HA solution and mixed under stirring at 30° C for 4 hrs. The different pH at the cross-linked HA solution were adjusted to pH 6.5-7.5 by addition of 1.00N NaOH or 1.0N HCl.

Please replace paragraph [0075] with the following amended paragraph:

[0075] Sodium hyaluronate (100 mg; 1 molar equivalent of hydroxyl group) was dissolved in 10 ml of distilled water. The 1% dry solid content of HA solution was formed under stirring at room temperature. ~~Following~~ Next, adjustment of the pH of the HA solution was performed by addition of 1.0 N NaOH or 1.0N HCl. These results were showed in the following table 3.

Please replace paragraph [0079] with the following amended paragraph:

[0079] Sodium alginate (200 mg; 2 molar equivalent of hydroxyl group) was dissolved in 20 ml of distilled water. The 1% dry solid content of sodium alginate solution was formed under stirring at room temperature. ~~Following~~ Next, adjustment of the pH of the HA solution was performed by addition of 1.0 N NaOH or 1.0N HCl.

Please replace paragraph [0082] with the following amended paragraph:

[0082] Chondroitin-6-sulfate (503 mg; 3 molar equivalent of hydroxyl group) was dissolved in 10 ml of distilled water. The 5% dry solid content of chondroitin-6-sulfate solution was formed under stirring at room temperature. ~~Following~~ Next, adjustment of the pH of the chondroitin-6-sulfate solution was performed by addition of 1.0 N NaOH or 1.0N HCl. These results were showed in the following table 5.

Please replace paragraph [0085] with the following amended paragraph:

[0085] Pectin (352 mg; 4 molar equivalent of hydroxyl group) was dissolved in 17.6 ml of distilled water. The 2% dry solid content of pectin solution was formed under stirring at room

temperature. ~~Following~~ Next, adjustment of the pH of the pectin solution was performed by addition of 1.0 N NaOH or 1.0N HCl. These results were showed in the following table 6.

Please replace paragraph [0088] with the following amended paragraph:

[0088] Sodium alginate (100 mg; 1 molar equivalent of hydroxyl group) was dissolved in 10 ml of distilled water. The 1% dry solid content of sodium alginate solution was formed under stirring at room temperature. ~~Following~~ Next, adjustment of the pH of the sodium alginate solution was performed by addition of 1.0 N NaOH or 1.0N HCl.

Please replace paragraph [0091] with the following amended paragraph:

[0091] Chondroitin-6-sulfate (503 mg; 3 molar equivalent of hydroxyl group) was dissolved in 10 ml of distilled water. The 5% dry solid content of chondroitin-6-sulfate solution was formed under stirring at room temperature. ~~Following~~ Next, adjustment of the pH of the chondroitin-6-sulfate solution was performed by addition of 1.0 N NaOH or 1.0N HCl. These results were showed in the following table 8.

Please replace paragraph [0094] with the following amended paragraph:

[0094] Sodium hyaluronate (100 mg; 1 molar equivalent of hydroxyl group) was dissolved in 10 ml of distilled water. The 1% dry solid content of HA solution was formed under stirring at room temperature. ~~Following~~ Next, adjustment of the pH of the HA solution was performed by addition of 1.0 N NaOH or 1.0N HCl. These results were showed in the following table 9.

Please replace paragraph [0097] with the following amended paragraph:

[0097] Sodium hyaluronate (100 mg; 1 molar equivalent of hydroxyl group) was dissolved in 10 ml of distilled water. The 1% dry solid content of HA solution was formed under stirring at room temperature. ~~Following~~ Next, adjustment of the pH of the HA solution was performed by addition of 1.0 N NaOH or 1.0N HCl. These results were showed in the following table 10.

Please replace paragraph [0117] with the following amended paragraph:

[0117] The resulting solution was cast into a mold of Teflon plate and allowed to dry under oven at 35° C to yield HA films, and then ~~immersed~~ **immersed** in the different organic solvent. The pH of the organic solvent was adjusted to pH 3.0 by adding 1.0N HCl solution. These results are showed in the table 16A.

Please replace paragraph [0125] with the following amended paragraph:

[0125] The pectin film was obtained from the same procedure as described above. The films were also ~~immersed~~ **immersed** in the organic solvent for the dissolubility test. These results are showed in the table (d) results showed that the pectin films were strong and insoluble at above 40/60 ratio of acetone/water solution.

Please replace paragraph [0138] with the following amended paragraph:

[0138] Powder of the CMC (200 mg; molecular weight  $1.4 \times 10^6$ ) was dissolved in 10 ml of distilled water. The 2% dry solid content of CMC solution was formed under stirring at room temperature. The resulting solution was cast into a mold and produced a porosity of the cross-linked polysaccharide by freeze-drying. The porosity of the CMC polysaccharide was ~~immersed~~ **immersed** in the EGDGE-containing organic solvent. The pH of the cross-linked solution was adjusted by adding 1.0N HCl or 1.0N NaOH solution. These results are showed in the table 20.

Please replace paragraph [0141] with the following amended paragraph:

[0141] Powder of the pectin (200 mg) was dissolved in 10 ml of distilled water. The 2% dry solid content of pectin solution was formed under stirring at room temperature. The resulting solution was pressed as a monofilament fiber of 50  $\mu\text{m}$ ~1 mm thickness by using the different size of syringe in a 95% alcohol solution. The pectin fibers were ~~immersed~~ **immersed** in the EGDGE-containing organic solvent. The pH of the organic solvent was adjusted by adding 1.0N HCl or 1.0 N NaOH. These results are showed in the table 21.



Please replace paragraph [0143] with the following amended paragraph:

[0143] Powder of the HA (200 mg) was dissolved in 10 ml of distilled water. The 2% dry solid content of pectin solution was formed under stirring at room temperature. The resulting solution was pressed as a monofilament fiber of 50  $\mu\text{m}$ ~1 mm thickness by using the different size of syringe in a 95% alcohol solution. The HA were ~~immeresed~~ **immersed** in the EGDGE-containing organic solvent. The pH of the organic solvent was adjusted by adding 1.0N HCl. These results are showed in the table 22.

Please replace paragraph [0145] with the following amended paragraph:

[0145] Powder of the CMC (200 mg; molecular weight  $1.4 \times 10^6$ ) was dissolved in 10 ml of distilled water. The 2% dry solid content of CMC solution was formed under stirring at room temperature. The resulting solution was cast into a mold and produced a porosity of the cross-linked polysaccharide by freeze-drying. The porosity of the CMC polysaccharide was ~~immeresed~~ **immersed** in the EGDGE-containing organic solvent. The pH of the cross-linked solution was adjusted by adding 1.0N HCl solution. These results are showed in the table 23. The porosity of the CMC polysaccharides were further cross-linked at the different temperature, the porosity of the CMC polysaccharides was dried under oven at 35° C.